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Annulohypoxylon (*Xylariales*) from western Paraná, Brazil

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Abstract — *Annulohypoxylon parvodiscum* sp. nov. is proposed, based on small ostiolar disc of *bovei*-type, olivaceous grey pigment, and ellipsoid ascospores (9–13 × 3–5 µm); and three other species of *Annulohypoxylon* are reported from western Paraná State, Brazil: *A. macrodiscum*, *A. nitens*, and *A. stygium*. A key for identification of studied taxa is provided.

Key words — *Ascomycota*, *Hypoxyloideae*, mycobiota, taxonomy, *Xylariaceae*

Introduction

Annulohypoxylon Y.M. Ju et al. was segregated from *Hypoxylon* Bull., based on morphological, chemotaxonomic and molecular data (Hsieh et al. 2005, Fournier et al. 2010). It covers taxa previously placed in *Hypoxylon* sect. *Annulata* J.H. Miller diagnosed by the presence of carbonaceous stromatic layer surrounding perithecia, projecting ostioles above the stromatic surface, with a conspicuous to inconspicuous annulate disc, and ascospores with dehiscent perispores (Ju & Rogers 1996). The genus comprises c. 40 species, of which sixteen are known from Brazil (Pereira 2015), most of them reported from the states of Bahia (Northeast) and Rio Grande do Sul (South). From Paraná State, only *A. stygium* has been reported (Meijer 2010).

In order to improve the knowledge on the diversity and geographical distribution of the *Xylariaceae* from western Paraná State, Brazil, a survey was undertaken (Cruz & Cortez 2015a, 2015b); here we present the results for the genus *Annulohypoxylon*.

Materials & methods

Specimens were collected from 2013 to 2014 in São Camilo State Park (abbreviated as PESC), municipality of Palotina, western Paraná State, Brazil (24°18′00″–19′30″S 53°53′30″–55′30″W) which comprises a relict of seasonal semi-deciduous forest (Atlantic Forest Domain; Kozera & Peluci 2015).

Morphology was examined according to Ju & Rogers (1996) and colors were based on Rayner (1970). Microscopic features were examined in 10% KOH, and measured in distilled water preparations; asci were mounted in Cotton Blue, except for the apical ring, examined in Melzer's reagent. Ascospores were analyzed under scanning electron microscope (SEM) Jeol JSM-6360LV, at the Center of Electron Microscopy of the Universidade Federal do Paraná, following Suwannasai et al. (2012). All specimens are preserved in the Herbarium, Department of Biodiversity, Universidade Federal Paraná, Palotina, Brazil (HCP), except for the *A. parvodiscum* holotype in the Herbarium, Department of Botany, Universidade Federal Paraná, Curitiba, Brazil (UPCB).

Taxonomy

Annulohypoxylon macrodiscum Jad. Pereira, J.D. Rogers & J.L. Bezerra, Mycologia 102: 250. 2010. Plates 1a–d, 3a

Stromata pulvinate to effused-pulvinate, 3–25 mm long × 5–14 mm broad × 0.5–1.5 mm thick; surface fuscous black (104) with brown vinaceous tone; blackish granules beneath surface and among perithecia, with KOH-diluted pigments, greenish olivaceous (90) or dull green (70); tissue below the perithecial layer, 0.3–0.5 mm thick. Perithecia spherical to obovoid, 0.6–1 × 0.5–1 mm, with perithecial mounds ¼ to ½ exposed. Ostioles conical-papillate, surrounded by a *truncatum*-type disc, 0.4–0.8 mm diam. Asci cylindrical, 114–121 × 5–7 µm; spore-bearing part 65–80 µm, stipe 40–50 µm; apical ring amyloid, inconspicuous, discoid, 0.5 × 1–1.3 µm. Ascospores brown to dark brown, ellipsoid-inequilateral, with narrowly rounded ends, 7–12 × 3.5–5 µm, with straight germ slit spore-length on the convex side; perispore dehiscent in KOH; epispore smooth; surface smooth under SEM.

Specimens examined: **BRAZIL. Paraná State**: Palotina, PESC, 13 Jun 2013, leg. K.S. Cruz 104 (HCP 583), 105 (HCP 584); 13 Dec 2013, leg. K.S. Cruz 179 (HCP 585), 180 (HCP 586).

Habitat and distribution: On rotting wood, in the board of forest trail. Known from Atlantic Forest, states of Bahia (Pereira et al. 2010) and Paraná.

Notes: *Annulohypoxylon macrodiscum* is diagnosed by the wide ostiolar disc and spherical to obovoid perithecia (Pereira et al. 2010). It is morphologically similar to *A. truncatum* (Schwein.) Y.M. Ju et al., but as discussed by Pereira et al. (2010), *A. truncatum* has smaller ostiolar disc (0.2–0.4 mm diam.) and perithecia (0.4–0.8 mm), which is also spherical (Ju & Rogers 1996). Ascospores of Paraná collections presented a slight variation in size, in comparison to type

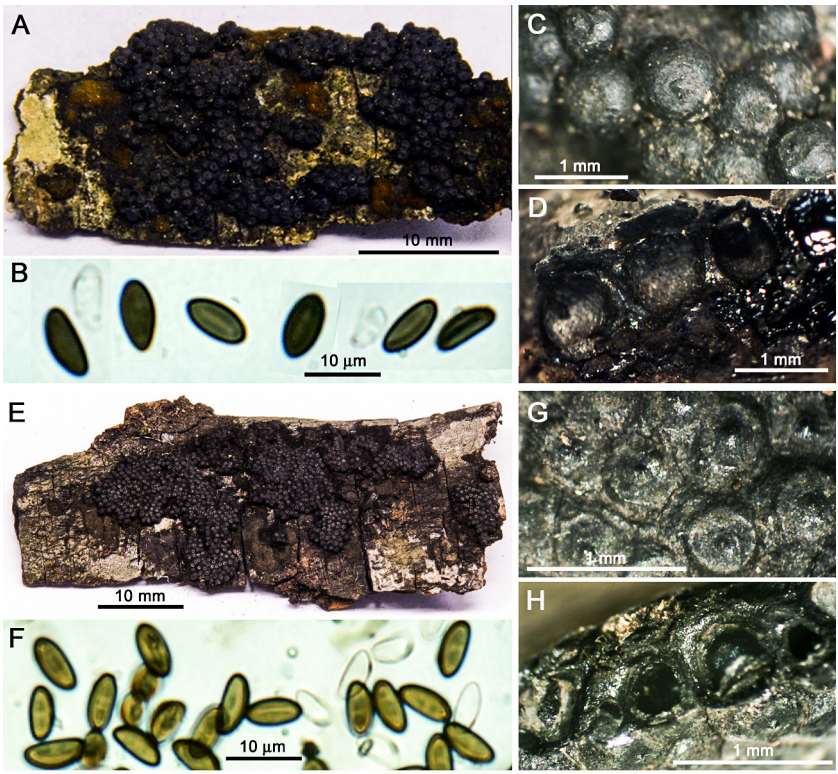

Plate 1. ***Annulohypoxylon macrodiscum***. A: stroma; B: ascospores; C: top view of disc; D: perithecia, sectioned. ***Annulohypoxylon nitens***. E: stroma; F: ascospores; G: top view of disc; H: perithecia, sectioned.

(8–11 × 4–5 µm; Pereira et al. 2010), but this is assumed to be intraspecific variation. The species was described from Northeastern Brazil (Pereira et al. 2010) and is now reported from Southern Brazil, but probably it is widespread throughout the Atlantic Forest biome.

Annulohypoxylon nitens (Ces.) Y.M. Ju, J.D. Rogers & H.M. Hsieh, Mycologia 97: 861. 2005. Plates 1e–h, 3b

Stromata glomerate to effused-pulvinate, 18–35 mm long × 5–12 mm broad × 0.5–1 mm thick, with perithecial mounds ¼ to ½ exposed; surface dark brown vinaceous (84) to fuscous black (104), with a white tissue covering the ostiolar disc; blackish granules beneath surface and among perithecia, with KOH-diluted pigments greenish olivaceous (90) or dull green (70); black tissue

below the perithecia inconspicuous, 0.2 mm thick. Perithecia spherical, 0.6–1 mm diam. Ostioles conical-papillate, encircled with a flattened *bovei*-type disc 0.3–0.5 mm diam. Asci 80–140 × 4–6 µm, spore-bearing parts 60–80 µm, stipe 21.5–59 µm, with apical ring amyloid, discoid, 0.5 × 1–1.5 µm. Ascospores light brown to brown, ellipsoid-inequilateral, with narrowly rounded ends, 6.5–9 × 3–5 µm, with straight germ slit spore-length on the convex side; perispore dehiscent in KOH, smooth; epispore smooth; surface smooth under SEM.

Specimens examined: **BRAZIL. Paraná State**: Palotina, PESC, 13 Jun 2013, leg. K.S. Cruz 100 (HCP 587); 10 Sep 2013, leg. K.S. Cruz 139 (HCP 588); 7 Oct 2013, leg. K.S. Cruz 173 (HCP 589); 18 Oct 213, leg. K.S. Cruz 184 (HCP 590), 186 (HCP 591); 5 Nov 2013, leg. K.S. Cruz 169 (HCP 592); 20 Jan 2014, leg. K.S. Cruz 196 (HCP 593), 198 (HCP 594); 25 Mar 2014, leg. K.S. Cruz 218 (HCP 595).

Habitat and distribution: On rotting wood, in the forest board. Known from the Americas and Asia (Ju & Rogers 1996, Pereira et al. 2015).

Notes: According to Ju & Rogers (1996), *A. nitens* presents spherical perithecia. However, our specimens showed spherical to obovoid perithecia, and were determined in the sense of Hladki & Romero (2009), who reported similar materials from Argentina. Similar species are *A. truncatum* and *A. moriforme* (Henn.) Y.M. Ju et al., which differ in the presence of an ostiolar disc of *truncatum*-type (Ju & Rogers 1996). Known from the state of Bahia, *A. nitens* is a new record from Paraná State, where it was one of the most common xylariaceous fungi sampled in PESC.

Annulohypoxylon parvodiscum K.S. Cruz & Cortez, **sp. nov.**

MycoBank MB 812745 Plates 2A–F, 3C

Differs from *Annulohypoxylon squamulosum* by its smooth vinaceous brown stromatal surface and larger ascospores.

Type: Brazil. Paraná State: Palotina, PESC, 13 June 2013, leg. K.S. Cruz 094 (**Holotype**, UPCB 81662; **isotype**, HCP 596).

Etymology: referring to the small (L., *parvus*) size of ostiolar disc (L., *discus*).

Stromata effused-pulvinate, 5–30 mm long × 3–8 mm broad × 0.5–0.8 mm thick; with inconspicuous perithecial mounds; surface brown vinaceous (84), smooth; blackish granules beneath surface and between perithecia, with KOH-diluted pigment olivaceous grey (121); tissue below the perithecial layer inconspicuous. Perithecia spherical to slightly obovoid, 0.3–0.5 × 0.2–0.4 mm diam. Ostioles slightly papillate, surrounded by a *bovei*-type disc, 0.1–0.2 mm diam. Asci cylindrical, 69–157.5 × 5–10 µm; spore-bearing part 68–103 µm, stipe 17.5–76 µm; apical ring amyloid, inconspicuous. Ascospores brown to dark brown, ellipsoid-inequilateral, with narrowly rounded ends, 9–13 × 3–5

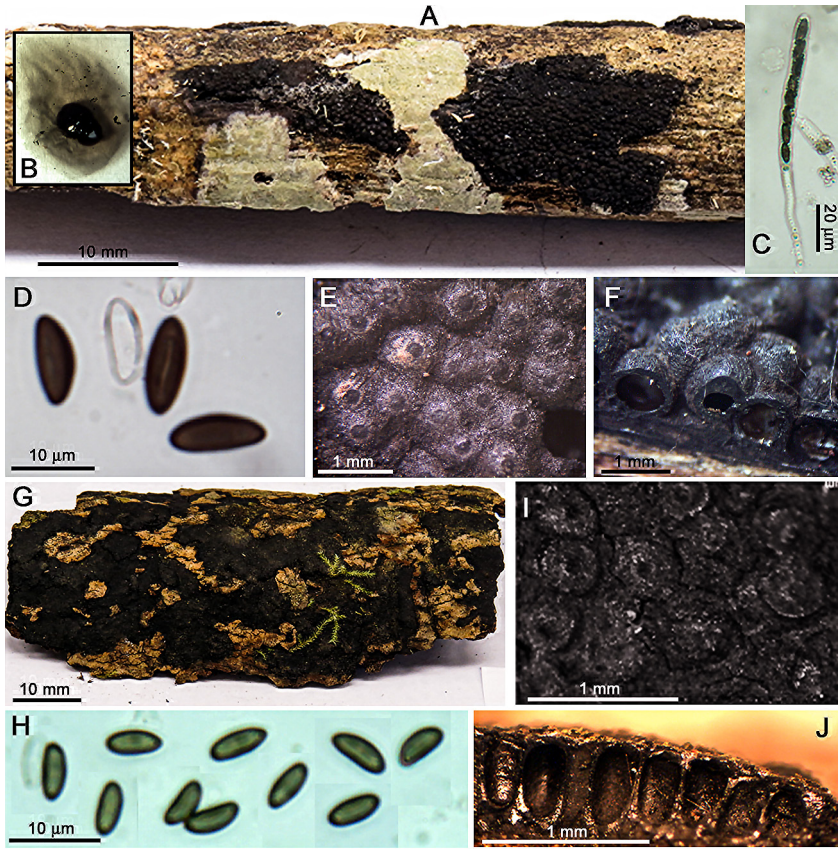

Plate 2. ***Annulohypoxylon parvodiscum*** (holotype). A: stroma; B: KOH-extractable pigment; C: asci; D: ascospores; E: top view of disc; F: perithecia, sectioned. ***Annulohypoxylon stygium***. G: stroma; H: ascospores; I: top view of disc; J: perithecia, sectioned.

µm, with straight germ slit spore-length, sometimes slightly sigmoid on the convex side; perispore dehiscent in KOH; episore smooth; surface smooth under SEM.

Additional specimens examined: **BRAZIL. Paraná State**: Palotina, PESC, 10 Sept. 2013, leg. K.S. Cruz 144 (HCP 597); 18 Oct. 213, leg. K.S. Cruz 185 (HCP 598).

Habitat and distribution: Growing on decayed angiosperm trunk, in the board of forest trail. Known only from type locality.

Notes: *Annulohypoxylon parvodiscum* is similar to *A. archeri* (Berk.) Y.M. Ju et al., *A. microcarpum* (Penz. & Sacc.) Y.M. Ju et al., and *A. squamulosum* (Y.M.

Ju et al.) Y.M. Ju et al. all species bearing reduced ostiolar disc (0.1–0.2 mm). However, in *A. archeri* and *A. microcarpum*, the disc is of *truncatum*-type (Ju & Rogers 1996). *Annulohypoxylon squamulosum,* which also has a *bovei*-type disc, differs by its rough stromatal surface and smaller ascospores (6.5–8.5 × 3–4 µm; Ju et al. 2004).

Annulohypoxylon stygium (Lév.) Y.M. Ju, J.D. Rogers & H.M. Hsieh, Mycologia 97: 861. 2005. Plates 2g–j, 3d

Stromata hemispherical to effused-pulvinate, 9–10 mm long × 5–7 mm broad × 0.3–0.4 mm thick, with inconspicuous perithecial mounds; surface fuscous black (104), with some reddish brown tone; reddish brown granules beneath surface and among perithecia, KOH-diluted pigments greenish olivaceous (90) or dull green (70); tissue below the perithecial layer inconspicuous. Perithecia obovoid, 0.3–0.4 × 0.15–0.25 mm diam. Ostioles conical-papillate, surrounded by a *truncatum*-type disc 0.1–0.2 mm diam. Asci cylindrical, 72–80 × 4–5 µm, spore-bearing part 56–62 µm, stipe 12–19 µm, apical ring amyloid, discoid, 0.5 × 1 µm. Ascospores light brown, ellipsoid-inequilateral, with narrowly rounded ends, 5–7 × 2.5–3 µm, with straight germ slit spore-length on flattened side; perispore dehiscent in KOH; epispore smooth; surface smooth under SEM.

Specimens examined: **BRAZIL. Paraná State**: Palotina, PESC, 16 May 2013, leg. K.S. Cruz 016 (HCP 578), 017 (HCP 579); 7 Oct 2013, leg. K.S. Cruz 174 (HCP 580), 183 (HCP 581); 5 Nov 2013, leg. K.S. Cruz 158 (HCP 582).

Habitat and distribution: On rotting wood, in the board of forest. Known from the Americas and Asia (Ju & Rogers 1996).

Notes: The small ascospores and ostiolar disc and green stromatic pigment are diagnostic features for *A. stygium* (Ju & Rogers 1996). It looks like the African *A. atroroseum* (J.D. Rogers) Y.M. Ju et al., which differs by its pinkish stromatic surface (Ju & Rogers 1996). In Brazil, *A. stygium* has been reported from Amazon and Atlantic Forest biomes, including Paraná State (Meijer 2006, 2010; Pereira 2015).

Key for the *Annulohypoxylon* species from PESC, Paraná, Brazil

1. Ostiolar disc *bovei*-type 2
1. Ostiolar disc *truncatum*-type 3
2. KOH-extractable pigments olivaceous grey, ostiolar disc 0.1–0.2 mm ..... ***A. parvodiscum***
2. KOH-extractable pigments olivaceous green, ostiolar disc 0.3–0.5 mm ... ***A. nitens***
3. Ostiolar disc 0.1–0.2 mm, ascospores 5–7 × 2.5–3 µm ..... ***A. stygium***
3. Ostiolar disc 0.4–0.8 mm, ascospores 7–12 × 3.5–5 µm ..... ***A. macrodiscum***

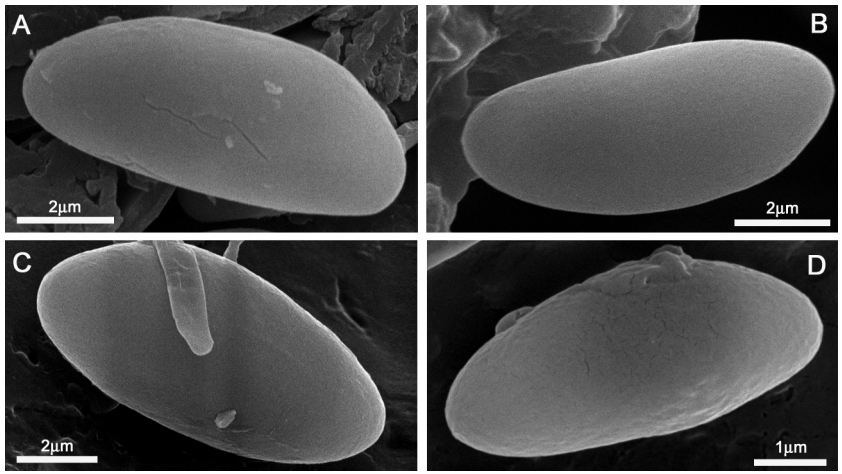

Plate 3. *Annulohypoxylon* ascospores (SEM).
A: *A. macrodiscum*. B: *A. nitens*. C: *A. parvodiscum*. D: *A. stygium*.

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